Influence of ¹³⁷Cs and ⁹⁰Sr on vegetative and generative organs of *Lepidium sativum* L. and *Tradescantia* clone 02

Danutė Marčiulionienė, Benedikta Lukšienė, Dalius Kiponas, Danguolė Montvydienė, <u>Gemir Maksimov</u>, Jūratė Darginavičienė, Virgilija Gavelienė

Abstract The impact of ¹³⁷Cs and ⁹⁰Sr activity concentrations from 0.4 to 400 kBq·L⁻¹ and from 1 to 200 kBq·L⁻¹, respectively, on seed germination of *Lepidium sativum* was insignificant; however, all concentrations of ¹³⁷Cs and 30 kBq·L⁻¹ concentration of ⁹⁰Sr slightly stimulated root growth. The accumulated ¹³⁷Cs and ⁹⁰Sr stimulated shoot and parenchyma cell growth; ¹³⁷Cs suppressed RNA-polymerase II activity, and ⁹⁰Sr, on the contrary, stimulated it. Different genotoxic effects on *Tradescantia* clone 02 stamen hair cells were observed with comparable ¹³⁷Cs and ⁹⁰Sr activity concentrations of ¹³⁷Cs from 0.001 to 1.3 kBq·L⁻¹ were more effective in *Tradescantia* clone 02 stamen hair cell reproduction, whereas the studied activity concentrations of ⁹⁰Sr (from 0.002 to 640 kBq·L⁻¹) induced more mutations.

Key words technogenic radionuclides • activity concentration • accumulation • *Tradescantia* clone BNL 02 • *Lepidium* sativum • RNA-polymerase II

D. Marčiulionienė[™], D. Kiponas, D. Montvydienė,
G. Maksimov, J. Darginavičienė, V. Gavelienė
Institute of Botany,
49 Žaliųjų ežerų Str., LT-08406, Vilnius, Lithuania,
Tel.: +370 5 2641790, Fax: +370 5 2729950,
E-mail: radeko@ar.fi.lt

B. LukšienėInsitute of Physics,231 Savanorių Ave., LT-02300, Vilnius, Lithuania

Received: 21 March 2006 Accepted: 30 August 2006

Introduction

Ionizing radiation can evoke various biological effects in plants, such as: inhibition or activation of DNA synthesis and synthesis of some metabolites, slackening or acceleration of cell division, growth suppression or stimulation, disturbance of ontogenesis phase dynamics, morphological changes of teratogenic origin, enhanced quantities of cells with increased numbers of chromosomal aberrations, increased number of individuals exhibiting chlorophyll mutations or new traits and reduced seed germination in progeny generations [11, 13, 18, 32, 35, 42]. It has been shown that in plants, radiocesium mostly accumulates (up to 25%) in the meristem (tissues where cells divide rapidly), which is relatively sensitive to ionizing radiation. Due to intensification of metabolic processes, large quantities of radionuclides can be accumulated in meristems, thus enhancing the internal irradiation doses [35, 37]. Radiostrontium mostly accumulates in plant tissues with dominating elongation growth and in those undergoing differentiation [18]. There are few papers dealing with plant response to the effect of incorporated ¹³⁷Cs [11]. Recently, only scarce studies on the impact of incorporated ¹³⁷Cs and ⁹⁰Sr on plants test-organisms have been performed [7, 8, 20, 23, 29].

Studies of the biological effects of the ionizing radiation of radionuclides are important for the assessment of radioecological condition of terrestrial and aquatic ecosystems in the surroundings of nuclear power stations, forecasting the ecotoxicological consequences of radionuclides released to the environment during accidents and managing the issues of ecological standards on the presence of radionuclides in the environment. The International Radioecology Union (IRU) points out that in the 21st century special attention should be given to the investigation of the effects of low ionizing radiation doses caused to biota, especially those due to the incorporation of radionuclides into organisms [5].

The objective of this work was to determine and to compare the toxic impact of technogenic radionuclides, ¹³⁷Cs and ⁹⁰Sr, incorporated in plant into the meristematic cells of root and parenchyma cells of the shoots and the genotoxic impact on stamen hair cells, as well as to estimate the effect of ¹³⁷Cs and ⁹⁰Sr on the initial stages of protein synthesis in plant shoot cells.

Materials and methods

¹³⁷Cs and ⁹⁰Sr model systems

Hydroponics system for model experiments was prepared from ¹³⁷CsCl and ⁹⁰SrCl₂ (Vsesojuznoe objedinenie "Izotop", Leningradskoe otdelenie, FSU). The ⁹⁰Sr isotope was in equilibrium with ⁹⁰Y. Initial 0.1 ml volume of each chloride was diluted 10^3 – 10^6 times or even more in order to obtain radionuclide activity concentration necessary for the study. In a separate variant of experimental series we used different radionuclide activity concentrations. Tests with *L. sativum* were carried out with ¹³⁷Cs (0.4, 4, 40, and 400 kBq·L⁻¹) and ⁹⁰Sr (1, 3, 30, and 200 kBq·L⁻¹) activity concentrations. The genotoxic studies of *Tradescantia* clone 02 were carried out at the following activity concentrations: ¹³⁷Cs, 0.001, 0.01, 0.13 and 1.3 kBq·L⁻¹, ⁹⁰Sr, 0.002, 0.02, 64 and 640 kBq·L⁻¹.

Test-organisms

Lepidium sativum L. Seed germination as well as root and shoot growth

The test was carried out following a modified Magone [24, 33] method. Briefly, 10 ml of lake water (as control) or a test aqueous solution of ¹³⁷Cs or ⁹⁰Sr was pipetted onto three layers of filter paper fitted into a 9-cm glass Petri dish. Twenty-five healthy looking *L. sativum* seeds of similar size were distributed evenly on the filter paper. The Petri dishes were placed in the darkness at $24 \pm 1^{\circ}$ C for 48 h. Afterwards, non-germinated seeds were counted, and root length was measured. The experimental set of each testing scheme involved 3 control dishes and 3 replications for each concentration activity of the radionuclides. The pH of lake water and test solutions with ¹³⁷Cs and ⁹⁰Sr was 7.5.

The level of toxic impact on *L. sativum* was assessed by the modified method of Wang [44]. According to the percent of root growth inhibition of 100-60%, 61-40%, 41-20% and lower than 19\%, the toxic impact of the tested sample solutions on *L. sativum* was classified as very strong, strong, moderate and weak, respectively. The tested concentration was considered non-toxic if the biological parameter of *L. sativum* did not statistically differ from the control, and was considered extremely toxic if the seed did not germinate.

In experiments on the influence of ¹³⁷Cs and ⁹⁰Sr on plant shoot growth, *L. sativum* were grown in plastic boxes ($110 \times 160 \times 6$ mm) with covers to avoid evaporation. Each box contained 65 ml of water solution and 470 mg (~160) of seeds that were evenly distributed on a glass plate, covered with filter paper. The seeds germinated at 24 ± 1°C for 24 h in the darkness, and the shoots were grown for 6 days at continuous light at a temperature of 23 ± 1°C.

The morphological anatomical studies included all *L. sativum* shoots. We measured length of each shoot (with straightened leaves) and the weight of all the shoots. A microscope was used to determine the length, width and area of parenchymal cells.

Tradescantia. Somatic mutations of stamen hair cells

Experiments with Tradescantia (Commelinaceae) clone 02 were performed applying modified Mericle and Mericle [30] and Osipova and Shevchenko [36] methods. Four stems of cuttings bearing in fluorescence were immersed in 200-ml glass flasks containing 150 ml of lake water (as control) or test ¹³⁷Cs or ⁹⁰Sr aqueous solution. The flasks were exposed to the 16-h light/8-h dark cycle for 14 days. The radionuclide genotoxic impact on Tradescantia clone 02 was evaluated by the number of somatic (colourless) mutations and morphological anomalies in the stamen hair (SH) as well as by the amount of non-viable SH (their number indicates lethality when a hair contains less than 12 cells), which were counted using a light microscope. The number of non-viable SH reflects the ability of cells to divide. In each case, approximately 8000-11,200 stamen hairs were counted. The number of somatic mutations in the Tradescantia clone 02 stamen hair system in the control (lake water) was $0.62\% \pm 0.05\%$, whereas non-viable SH were not observed. The degree of ¹³⁷Cs and ⁹⁰Sr genotoxicity was evaluated according to the methods suggested by Marčiulionienė and coauthors [26]. A slight genotoxic effect on the Tradescantia clone 02 SH system is observed when the number of somatic mutations and morphologic anomalies do not exceed 1%, and ability of stamen hair cells to divide is 100% (e.g. no non-viable SH). The medium effect is observed when the number of somatic mutations and morphologic anomalies is between 1-4%, and ability of SH cells to divide reaches 60%. Strong genotoxic effect is characterized by the number of somatic mutations and morphologic anomalies exceeding 4%, and ability of SH cells to divide less than 60%.

RNA-polymerase II activity

Cell nuclei from the shoots of *Lepidium sativum* plants were isolated using the conventional methods [39] with modifications [31]. The isolated nuclei were incubated in a medium suitable to induce RNA-polymerase II activity [21, 41]. The incubation medium contained

triphosphates GTP, UTP, CTP (all disodium salt, Fluka Chemie AG, Switzerland) and ¹⁴C-ATP (0.1 mM, spec. act. 3.1 MBq/g, Amersham Pharmacia Biotech, UK), Tris (Sigma-Aldrich Chemie GmbH, Steinheim, Germany)-HCl (P.A. Czech Republic) pH 7.6. The enzyme's activity was stopped after 40 min of incubation at +37°C by adding cooled trichloroacetic acid (final conc. 3%, Lach:Ner, Neratovice, Czech Republic). The residue was collected on membrane filters (\emptyset 2.5 µm; Pragopor, PRAGOCHEMA, Czech Republic) and washed with trichloroacetic acid and ethanol. A scintillation counter (Beckman LS 1801, USA) was used to measure the activity of incorporated ¹⁴C-ATP. The Bradford method [4] was used to determine the protein content.

¹³⁷Cs and ⁹⁰Sr activity concentration measurements

¹³⁷Cs activity concentrations in the solution and in dry plant biomass were assessed by the method of gammaspectral analysis. In order to assess ¹³⁷Cs activity in the solution, 3 ml of different activity solutions were placed in each of the vials of standard geometry. ¹³⁷Cs activity concentration in the small-volume samples was measured with a gamma spectrometer interfaced with a p-type high purity germanium (HPGe) detector (GWL-series), equipped with a well 40 mm deep and 16 mm in diameter, made by EG&G ORTEC. The relative efficiency of the detector was 17% (for ¹³⁷Cs 661.7 keV radiation). The measurement uncertainty did not exceed 6%, with a statistic error not larger than 1% [17].

 50 Sr activity in the solution and plants was measured using a low background device UMF-1500 M (detector BT-13, registration efficiency 23% – 0.06 cps) (KIP, Tallinn, Estonia, FSU).

Statistical analysis

The data presented below are the arithmetical means of 2–3 experiments for which the standard errors of

estimations were calculated. Standard errors did not exceed 5% for all data. A statistically significant difference between experimental and control samples was assessed by the t-test (at p < 0.05) using Statgraphics Plus Version 2.1 program (Statistical Graphics Corp., Herndon, USA).

Results and discussion

In reported laboratory experiments, the influence of variable ¹³⁷Cs and ⁹⁰Sr (prepared from ¹³⁷CsCl and ⁹⁰SrCl₂) radioactivity concentrations upon the vegetative and generative organs of plants in the hydroponic medium was investigated.

Several experiments have been described concerning the effects of Cs⁺ from CsCl on physiological processes in cress seedlings. Cesium (3–4 mM solutions) seems to disturb water uptake and tissue hydration in cress [9]; cesium concentrations of 2–4 mM also evoke osmotic stress in *Arabidopsis* [46]. Millimolar concentrations of cesium chloride induce a strong osmotic stress in the cress seedlings. This is evident from the decrease of water uptake and hydration of the tissues [43].

In the present study hydroponic medium was contaminated with ¹³⁷Cs and ⁹⁰Sr, which were prepared from 0.1 ml ¹³⁷CsCl and ⁹⁰SrCl₂ by dilution 10^3-10^6 or more times. We assume, therefore, that non-radioactive cesium and strontium could not affect the studied *Lepidium sativum* and *Tradescantia* indices.

Investigation of the toxic impact of different ¹³⁷Cs activity concentrations (from 0.4 to 400 kBq·L⁻¹) on *L. sativum* seed germination and root growth revealed that after 2 days the seed germination did not statistically differ from the controls. However, this radionuclide slightly stimulated (11–12%) root growth (Fig. 1). Seed germination of *L. sativum* in the tested activity concentration (1–200 kBq·L⁻¹) of ⁹⁰Sr did not statistically differ from controls. However, ⁹⁰Sr activity in the range 1–30 kBq·L⁻¹ after 2 days induced a statistically significant (8–12%) inhibition of root growth; the highest used ⁹⁰Sr activity concentration (200 kBq·L⁻¹),



Fig. 1. Impact of incorporated ¹³⁷Cs and ⁹⁰Sr on seed germination and root growth of *Lepidium sativum* (after 2 days) depending on radionuclide activity concentration.



Fig. 2. Effect of ¹³⁷Cs and ⁹⁰Sr on shoot growth of *Lepidium sativum* (after 7 days) depending on radionuclide activity concentration.

on the contrary, stimulated the root growth (14%) as compared to control (Fig. 1). Analysis of ¹³⁷Cs effect on *L. sativum* shoot growth showed that after 7 days of exposure to the solution containing ¹³⁷Cs, shoot height and particularly weight had been stimulated only by the highest ¹³⁷Cs concentrations (Fig. 2). In many cases shoot parenchymal cell length and width stimulation was observed, which was most pronounced when ¹³⁷Cs activity concentration was 40 kBq·L⁻¹ (Fig. 3).

Analysis of ⁹⁰Sr effect on *L. sativum* plant shoot growth revealed that after 7 days of exposure to ⁹⁰Sr solution, the stimulation of both shoot height and weight was most intensive at a ⁹⁰Sr activity concentration of 30 kBq·L⁻¹ (Fig. 2). The same ⁹⁰Sr activity concentration stimulated shoot parenchymal cell length and width much more significantly than other studied concentrations (Fig. 3).

Out of numerous possible indices of cell functional state RNA-polymerase II activity was selected. It is directly connected with the process of RNA synthesis, and, therefore, it is informative of the primary stages of protein synthesis in the nucleus. RNA-polymerase II activity was controlled in a model of isolated nuclei in the RNA synthesis system by the amount of tracer ¹⁴C-ATP which is dependent on α -amanitine (mRNA synthesis inhibitor) and in normal growth conditions correlates with cell growth intensity [10].

The model system of the isolated nuclei was formed of the nuclei isolated from cells of *L. sativum* shoots grown in a medium containing ¹³⁷Cs. All studied ¹³⁷Cs activity concentrations suppressed the RNApolymerase II activity (Fig. 4). It was mostly pronounced at 400 kBq·L^{-1 137}Cs activity concentration (by 66% less than in control). Recalculation of the obtained results for the number of shoots used in the test did not affect the trend observed for this radionuclide. Variable ⁹⁰Sr activity concentrations stimulated RNApolymerase II activity of *L. sativum* shoots (Fig. 4).

It is known that tissues of plant generative organs are more sensitive to the impact of ionizing radiation than the tissues of vegetative organs [11, 38]. Cytogenetic effects are among the well-defined criteria for estimation of the impact of ionizing radiation upon biota [38]. The present investigation of the genotoxic



--- Control ---- Shoots parenchymal cell length ---- Shoots parenchymal cell width

Fig. 3. Effect of ¹³⁷Cs and ⁹⁰Sr on parenchymal cell parameters of *Lepidium sativum* (after 7 days) depending on radionuclide activity concentration.



Fig. 4. Influence of ¹³⁷Cs and ⁹⁰Sr on the activity of RNA-polymerase II in the model RNA-synthesis system of nuclei isolated from *Lepidium sativum* cells depending on radionuclide activity concentration (standard errors did not exceed 5%).

impact of the radionuclides on *Tradescantia* clone 02 showed that on the 14th day of treatment the lowest activity concentration of ¹³⁷Cs caused 1.4% of somatic mutations and morphological anomalies and decreased the ability of cell division by 19%, while the effects of the lowest activity concentration of ⁹⁰Sr on these parameters were 1.8% and 28%, respectively (Fig. 5). The increase in the activity concentration of ¹³⁷Cs (from 0.001 to 1.3 kBq·L⁻¹) and ⁹⁰Sr (from 0.002 to 640 kBq·L⁻¹) caused a decrease in the cell division ability of stamen hair cells of *Tradescantia* clone 02, whereas the number of somatic mutations and morphological anomalies showed no significant change (Fig. 5).

The impact of comparable activity concentrations of ¹³⁷Cs and ⁹⁰Sr on the root growth of *L. sativum* was different. The root growth of *L. sativum* was slightly stimulated by all the studied activity concentrations of ¹³⁷Cs and only by 30 kBq·L⁻¹ activity concentration of ⁹⁰Sr (Table 1, Fig. 1). Lower ⁹⁰Sr activity concentrations slightly inhibited the root growth. Thus, *L. sativum* root growth dependence on the studied activity concentration of ¹³⁷Cs and ⁹⁰Sr was observed, but for seed germination no such dependence was found. Shevchenko with co-authors [38] states that the alterations of meristematic tissues exposed to radiation are related to the morphological alterations of cells, and they can be important for the subsequent plant growth. The reasons for the difference of radiosensitivity of plant meristematic tissues may be linked to metabolism disturbances [11, 38]. ¹³⁷Cs and ⁹⁰Sr treatment slightly increased shoot

¹³⁷Cs and ⁹⁰Sr treatment slightly increased shoot height and weight of *L. sativum* as well as parenchymal cell length and width (Table 1). The increase was mostly evident when the activity concentration of ¹³⁷Cs was 40 kBq·L⁻¹ and that of ⁹⁰Sr – 30 kBq·L⁻¹; however, the stimulating effect of ⁹⁰Sr was much stronger than that of ¹³⁷Cs. At the activity concentration of ¹³⁷Cs, 400 kBq·L⁻¹ and of ⁹⁰Sr, 200 kBq·L⁻¹, the increase in shoot height and weight as well as in parenchymal cell length and width was much slighter, and these parameters did not statistically differ from the control (Table 1). Naidich [34] stated that the analysis of morphometric indices which characterize the viability of seedlings in the early stages of plant development



---- Somatic mutations and morphologic anomalies

-O- Cell division ability

Fig. 5. Impact of ¹³⁷Cs and ⁹⁰Sr on *Tradescantia* clone 02 stamen hair cells (after 14 days) depending on radionuclide activity concentration.

2	
clone 0	
descantia	
and Trae	
tivum L.	
idium sa	
n on <i>Lep</i>	
centratio	
ivity con	
d ⁹⁰ Sr act	
¹³⁷ Cs an	
variable	
Effects of	
e 1. I	

Table 1	. Effects of variab	ble 137 Cs and 90 Sr i	activity concent	ration on <i>Lepidiu</i>	<i>m sativum</i> L. a	nd Tradescanti	a clone 02				
Radio- nuclide	Radiation, energy, intensity	Lepidium sativu	<i>m</i> L.							<i>Tradescantia</i> clon (after 14 days)	e 02
	ALICHIM	Initial activity	Roots (after	2 davs)	Shoots (after	7 davs)				Initial activity	Genotoxic
		concentration in aqueous solution [kBq·L ⁻¹]	Seed germination	Root length	Height	Weight	Parenchyma Length	l cell Width	Activity of RNA-poly- merase II	concentration in aqueous solution [kBq·L ⁻¹]	effect
137 Cs	β ⁻ , 0.51 MeV, 92 <i>0</i> / ₆	0.4	no effect	slight stimulation	no effect	no effect	no effect	no effect	strong inhibition	0.001	moderate
	β ⁻ , 1.17 MeV, 8%	4	no effect	slight stimulation	no effect	no effect	no effect	slight stimulation	strong inhibition	0.01	strong
137m Ba	γ^{-} , 0.661 MeV	40	no effect	slight stimulation	slight stimulation	slight stimulation	no effect	slight stimulation	strong inhihition	0.13	strong
		400	no effect	slight stimulation	no effect	slight stimulation	no effect	no effect	strong inhibition	1.3	strong
90 Sr	β ⁻ , 0.535 MeV	1	no effect	slight inhibition	no effect	slight stimulation	no effect	no effect	slight stimulation	0.002	strong
λ_{06}	β ⁻ , 2.24 MeV, ~100%	3	no effect	slight inhibition	no effect	slight stimulation	no effect	no effect	slight stimulation	0.02	strong
	γ , 1.75 MeV	30	no effect	slight inhibition	slight stimulation	strong stimulation	slight stimulation	slight stimulation	strong stimulation	64	strong
		200	no effect	slight stimulation	slight stimulation	no effect	no effect	no effect	moderate stimulation	640	strong

showed that stimulation of shoot growth by ionizing radiation was observed in parallel with enhanced cytogenetic damages. Consequently, the stimulation of plant growth cannot be considered as useful or at least not harmful response of plant to the impact of ionizing radiation.

¹³⁷Cs, at the activity concentrations of 0.4–400 kBq·L⁻¹, stimulated the growth of *L. sativum* root and shoots and suppressed RNA-polymerase II activity (Table 1). Growth reflects almost all processes that take place in the cell; therefore, for the elucidation of the toxic effects, the internal processes on which growth change is based are relevant. The effect of ¹³⁷Čs with respect to cells is stressogenic; therefore, it may influence the processes taking place in the cell nucleus. Studies on RNA-polymerase II activity in isolated nuclei showed that all tested ¹³⁷Cs activity concentrations (from 0.4 to 400 kBq·L⁻¹) inhibited the functioning of RNApolymerase II. Although ¹³⁷Cs at test concentrations stimulated plant growth, it suppressed the initial process of protein synthesis, that is, transcription. The process of enhanced growth in plants can be caused by an enhanced functioning of the cell systems not related to the processes controlled by RNA-polymerase II, such as intensified water absorption, stock reserves of seeds and growing cells, etc.

The highest stimulation by ⁹⁰Sr of RNA-polymerase II activity as well as of the morphological and anatomical indices of shoot growth was observed at a 30 kBq·L⁻¹ concentration of this radionuclide (Table 1). The obtained results correspond to the correlations of physiological processes in plants: under an enhanced activity of cell metabolism and nuclear processes, cell growth is accelerated and thus, the morphological and anatomical indices of shoot growth increase.

The stimulating effect of radionuclides can cause morphogenetic changes in plants manifested in early developmental stages [26, 30]. Morphological changes in plants were observed after the Chernobyl NPP accident in the 30 km exclusion zone around the NPP [14]. Plant morphological changes due to damaged reproductive organs can also decrease germination of ripe seeds. It has been found that toxicants at concentrations not exceeding the levels producing toxic effects can stimulate the plant metabolism as well as growth processes in plants and their cells [1, 7]. Nevertheless, the plant enzyme activity can be disturbed by metabolic products, and the degree of injuries depends on the intensity of metabolism [1, 6]. Geraskin with co-authors [12] noted that the storage and reprocessing of low and intermediate activity waste caused additional environmental contamination, which induced cytogenetic disturbances of both vegetative and reproductive organs in Scotch pine. In fact, exposure to ionizing radiation causes some alterations in plants populations such as chromosomal aberrations, visible mutations, biochemical mutations, changes in genetic structure of the population, extinction of sensitive species, and, at last, degradation of the ecosystem [38].

At comparable ¹³⁷Cs and ⁹⁰Sr activity concentrations, the genotoxic effect on *Tradescantia* stamen hair cells was different (Table 1, Fig. 5). ¹³⁷Cs at all studied activity concentrations (from 0.001 up to 1.3 kBq·L⁻¹) was more effective on *Tradescantia* stamen hair cell division, whereas ⁹⁰Sr at the concentrations from 0.002 to 640 kBq·L⁻¹ induced more mutations (Table 1, Fig. 5). Similar results were obtained by other authors who studied radionuclide impact on test-organisms [22].

Different impact of the studied ¹³⁷Cs and ⁹⁰Sr activity concentrations on L. sativum root meristemic cells and shoot parenchyma cells, on the activity of RNApolymerase II and on the Tradescantia SH system (Table 1) can be explained by different metabolism of these radionuclides in plant. The transport pathway and distribution of ¹³⁷Cs and ⁹⁰Sr in plants are different [3, 4, 11, 45] because their stable chemical analogs are macro elements K and Ca, respectively. The physiological similarities of Cs and K and Sr and Ca are frequently indicated in radioecological studies [3, 4, 45, 47]. The highest amounts of ⁹⁰Sr in the plant cell are localized in chloroplasts, while ¹³⁷Cs distributes evenly in cell protoplasm [15, 25]. Accumulation of ¹³⁷Cs in cell wall, depending on plant species, is from 2 to 7 times smaller than that of ⁹⁰Sr. However, the release of ¹³⁷Cs from cell wall to protoplasm is higher (10-20%) than that of 90 Sr (3–10%) [25]. The distribution of these radionuclides in organs and tissues of plant also is different [40]. ¹³⁷Cs in plants accumulates mostly in the zones of cell division and active metabolism (e.g., in plant meristem and young tissues) [11], whereas ⁹⁰Sr is mostly accumulated in plant tissues with dominating elongation growth and in the young tissues of stem and leaves [18]. ⁹⁰Sr activity concentration in the aboveground part of 7 species of plants from different biotopes (forest, grassland and wetland) was from 2 to 12 times higher than that in the underground part of these plants [16]. The distribution of ¹³⁷Cs activity concentration in the same plant species was different. The activity concentration of this radionuclide in some species was higher in the aboveground part of plant, while in other species it was higher in the underground part of plant [27]. Comparison of the ¹³⁷Cs activity concentration in L. sativum roots and aboveground part demonstrated that after 7 days ¹³⁷Cs activity concentration was 5 times higher in the roots than in the aboveground part of plant when L. sativum was cultivated hydroponically, and during the experiment the roots were incubated in lake water solution of $40 \text{ kBq} \cdot \text{L}^{-1 \text{ 137}}$ Cs activity concentration [28]. Despite the similarity in chemical activity of Sr and alkaline metals, the effects of comparable activity concentration of 137 Cs and 90 Sr on the *L*. sativum root growth, shoot height and weight as well as parenchyma cell length and width, and RNA-polymerase II activity were different. The physiological similarity of cesium and potassium is frequently indicated in radioecological and physiological studies [11]. However, in the studies [11, 43] differences in cesium and potassium effects on some physiological parameters of L. sativum were determined. These distinctions could be due to the much higher atomic weight and ionic radius of cesium than those of potassium [11]. Basing on these statements, the influence of cesium on vegetative and generative organs of test-organisms should be stronger than that of strontium because of its higher atomic weight (133 against 87.6) and ionic radius (167 pm against 118 pm) [2, 19]. The summarized effects of variable ^{137}Cs and ⁹⁰Sr activity concentrations on *L. sativum* and *Tradescantia* clone 02 (Table 1) did not show much stronger impact of ¹³⁷Cs as compared to ⁹⁰Sr. Such differences could be discussed considering different properties of these elements as radioisotopes. ⁹⁰Sr+⁹⁰Y β-radiation is significantly higher than ¹³⁷Cs β-radiation (Table 1) and emitted γ-rays of ^{137m}Ba are softer (0.061 MeV) than those occurring in the ⁹⁰Y decay scheme (1.75 MeV) [2]. It can be concluded that the observed ¹³⁷Cs and ⁹⁰Sr influence on the test-organisms corroborates our statement that ¹³⁷CsCl and ⁹⁰SrCl₂ used in our experiments had no impact as stable cesium and strontium.

Conclusions

All the studied ¹³⁷Cs activity concentrations $(0.4-400 \text{ kBq}\cdot\text{L}^{-1})$ have been found to induce the slight stimulation of *L. sativum* root growth. Among all ⁹⁰Sr activity concentrations (1–200 kBq·L⁻¹) only the highest one increased *L. sativum* root growth. All other ⁹⁰Sr activity concentrations slightly inhibited root growth.

The highest ¹³⁷Cs activity concentrations (40–400 kBq·L⁻¹) slightly induced the shoot growth of *L. sativum*. ⁹⁰Sr also stimulated shoot growth, the stimulation being much more significant than in case of ¹³⁷Cs. All the studied ⁹⁰Sr activity concentrations slightly stimulated shoot growth, while 30 kBq·L⁻¹ activity concentration of this radionuclide strongly stimulated *L. sativum* shoot growth.

All studied ¹³⁷Cs activity concentrations strongly suppressed RNA-polymerase II activity, whereas ⁹⁰Sr, particularly at a 30 kBq·L⁻¹ activity concentration, strongly activated RNA-polymerase II.

All tested ¹³⁷Cs and ⁹⁰Sr activity concentrations caused a strong genotoxic effect in *Tradescantia* clone 02 stamen hair system. ¹³⁷Cs at all studied activity concentrations (from 0.001 up to 1.3 kBq·L⁻¹) affected to a greater extent all divisions in *Tradescantia* stamen hair, whereas ⁹⁰Sr at the concentrations from 0.002 to 640 kBq·L⁻¹ induced more mutations.

Different effect of ¹³⁷Cs and ⁹⁰Sr on the growth of plant vegetative organs (roots and shoots), on RNApolymerase II activity in RNA synthesis system of isolated plant cell nuclei and on the cells of a generative organ (flower) stamen hair cells can be explained by different metabolism of ¹³⁷Cs and ⁹⁰Sr, as stable chemical analogues of K and Ca, respectively, in the plant cells. This predetermines their variable accumulation in separate plant organs and their different distribution in plant cells and tissues.

Acknowledgment The study was supported by the Lithuanian State Science and Studies Foundation.

References

 Adelman R, Saul RL, Ames BN (1988) Oxidative damage to DNA: relation to species metabolic rate and life span. Proc Natl Acad Sci USA 85:2706–2708

- 2. Arcimovich LA (ed) (1963) The nuclear handbook. Governmental Publishers of Physico-Mathematical Literature, Moscow (in Russian)
- 3. Bauer CS, Plieth C, Bethmann B *et al.* (1998) Strontiuminduced repetitive calcium spikes in a unicellular green alga. Plant Physiol 117:545–557
- 4. Bradford MM (1976) A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72;1/2:249–254
- Brechignac F, Polikarpov G, Oughton DH et al. (2003) Protection of the environment in the XXI century: radiation protection of the biosphere including humankind. Statement of the International Union of Radioecology. J Environ Radioact 70;3:155–159
- 6. Britt AB (1996) DNA damage and repair in plants. Annu Rev Plant Physiol Plant Mol Biol 47:75–100
- Butkus D, Andriulaitytė I, Lukšienė B, Druteikienė R (2003) Peculiarities of radionuclide transfer to plants. J Environ Engin Landscape Manag 9;3:93–99
- Bystrzejewska-Piotrowska G, Drożdż A, Stęborowski R (2005) Resistance of heather plants (*Calluna vulgaris* L.) to cesium toxicity. Nukleonika 50;1:31–35
- 9. Bystrzejewska-Piotrowska G, Urban PL (2003) Accumulation of caesium in leaves of *Lepidium sativum* and its influence on photosynthesis and transpiration. Acta Biol Cracov Bot 45;2:131–137
- Darginavičienė J, Zemėnas J, Banienė J (2000) The effect of indole-3-acetic acid – protein complexes of wheat coleoptile plasmalemma from dividing and elongating cells on RNA-polymerase II activity. Biologija 2:134–136
- Evseeva TI, Geraskin SA, Khramova ES (2001) Cytogenetic effects of separate and combined action of ²³²Th and Cd nitrates on *Allium cepa* root tip cell. Citologija 43;8:803–808 (in Russian)
- 12. Geraskin SA, Zimina LM, Dikarev VG *et al.* (2000) Comparative analysis of anthropogenic combination in the region of radioactive waste reprocessing and storing plant arranged in the 30-km zone of the ChNPP, using bioindicative methods. Ekologija 4:300–303 (in Russian)
- Goncharenko GG, Surkov AA (2003) Change in mutation processes in environmental coniferous population in southern Belarus. In: Chudakova VA (ed) Proc on IV Int Symp on Actual Problems of Dosimetry, 7–8 October 2003, Minsk, Belarus, pp 38–39 (in Russian)
- Grodsinsky DM (ed) (2001) 15 Years of the Chernobyl catastrophe. Bulletin of NCRPU, Kiev, Ukraine, pp 1–136
- Grodsinsky DM, Kolomijec K, Kutlakhmedov J, Bulakh A, Dmitriev A, Khomljak M (1991) An anomaly of anthropogenic radionuclides and plants. Lybid, Kiev (in Russian)
- Gudelienė I, Marčiulionienė D, Petrošius R (2006) General regularities of ⁹⁰Sr distribution in system soil – plant under natural condition. In: Kundas SP, Okeanov AE, Pozniak SS (eds) Proc Int Conf Sakharov Readings 2006: Environmental problems of the XXI century, 18–19 May 2006, Minsk, Belarus 1:3–6
- Gudelis A, Remeikis V, Plukis A, Lukauskas D (2000) Efficiency calibration of HPGe detectors for measuring environmental samples. Environ Chem Phys 3/4;22:117–125
- Gudkov IN (2001) Peculiarities of the formation of internal exposure doses from radionuclides and radiobiological effects on plants. In: Taskaev AI, Kudiasheva AG, Ermakova OV, Popova ON (eds) Proc Int Conf on Biological Effects of Low Dose Ionizing Radiation and Radioactive Pollutions on Environment, 17–21 September 2001, Syktyvkar, Russia, pp 192–193 (in Russian)

- Jasinskis V, Matulis B, Pažarauskas E, Rinkevičienė ED, Zelionkaitė V, Žarnauskas AJ (1995) General and inorganic chemistry. Publishers of Science and Encyclopedias, Vilnius (in Lithuanian)
- Korogodina V, Bamblevsky V, Grishina I et al. (2004) Evaluation of the consequences of stress factors on plants seeds growing in the 30-km zone of Balakovo NPP. Radiacionnaja biologija, Radioekologija 44;1:81–88 (in Russian)
- Kulaeva O, Selivankina S, Nikolaeva N, Nichiporovich A (1979) Activation of RNA-polymerase II from isolated cell nuclei and chloroplast by citokinine. Plant Physiol 26:1016–1027 (in Russian)
- 22. Lamb T, Bickham JW, Lyne TB, Gibbons JW (1995) The slider turtle as an environmental sentinel: multiple tissues assays using flow cytometric analysis. Ecotoxicol 4:5–13
- 23. Lukšienė B, Butkus D, Druteikienė R (2004) Peculiarities of gaseous ⁸⁵Kr and ionic state ¹³⁷Cs accumulation in plants. In: Anke M *et al.* (eds) Proc of the 22th Workshop of Macro and Trace Elements. Agricultural, biological, environmental, nutritional and medical importance of macro, trace and ultra trace elements, 24–25 September 2004, Jena, Germany 1:830–835
- Magone I (1989) Bioindication of toxicity of transport emission. In: Kachalova OL, Lapinia IM, Melecis VP (eds) The impact of highway emission on natural environment. Zinatna, Riga, pp 108–116 (in Russian)
- Marčiulioniene D (1994) Radionuclides interaction with hydrophytes in freshwater ecosystems. Dissertatio Doctoralis ad Habilitationem. Institute of Ecology, Vilnius (in Russian)
- Marčiulionienė D, Dušauskiene-Duž R, Motiejunienė E, Švobienė R (1992) Radiochemoecological situation in Lake Druksiai – cooling water reservoir of the Ignalina NPP. Academia, Vilnius (in Russian)
- 27. Marčiulionienė D, Kiponas D, Hansen D (2001) Accumulation of technogenic radionuclides in the environment of Ignalina NPP. Ekologija 1:52–9 (in Lithuanian)
- Marčiulionienė D, Kiponas D, Lukšienė B (2005) Pecularities of ¹³⁷Cs activity distribution in the aquatic solution – solid phase – plant system. Ekologija 4:20–27
- Marčiulionienė D, Montvydienė D, Kiponas D, Dušauskiene-Duž R, Lukšienė B (2003) Genotoxic impact of ionizing radiation at low exposure doses of technogenic radionuclides accumulated in plants. Environ Chem Phys 4;25:218–227
- Mericle IW, Mericle RP (1967) Genetic nature of somatic mutations for flower color in *Tradescantia*, clon 02. Radiat Botany 7;6:449–464
- Merkys A, Darginavičienė J (1997) Plant gravitropic response. Adv Space Biol Med 6:213–230
- Minouflet M, Ayrault S, Badot PM, Cotelle S, Ferard JF (2005) Assessment of the genotoxicity of ¹³⁷Cs radiation using *Vicia*-micronucleus, *Tradescantia*-micronucleus and *Tradescantia*-stamen-hair mutation bioassays. J Environ Radioact 81;2/3:143–153
- Montvydienė D, Marčiulionienė D (2004) Assessment of toxic interactions of heavy metals in a multicomponents mixture using *Lepidium sativum* and *Spirodela polyrrhiza*. Environ Toxicol 19;4:351–358

- Naidich BI (1999) Fundamental march in radiobiology in 1998. Radiacionnaja biologija, Radioekologija 36;2/3:360–367 (in Russian)
- 35. Nesterov EB, Dikarev VG, Dikareva NS, Geraskin SA (2001) Cytogenetic effects of various doses rate of ionizing radiation on the meristemic tissue of barley roots. In: Taskaev AI, Kudiasheva AG, Ermakova OV, Popova ON (eds) Proc Int Conf on Biological Effects of Low Dose Ionizing Radiation and Radioactive Pollutions on Environment, 17–21 September 2001, Syktyvkar, Russia, pp 228–229 (in Russian)
- 36. Osipova RG, Schevchenko VA (1984) The use *Tradescantia* (clones 02 and 4430) in studies on radiation and chemical mutagenesis. J Gen Biol 45:226–232 (in Russian)
- 37. Shershunova VI, Khomichenko AA, Prilepova NV, Aniskina MV (2001) Influence of low radiation doses on *Tradescantia* clone 02 and *Arabidopssis thaliana* (L). In: Taskaev AI, Kudiasheva AG, Ermakova OV, Popova ON (eds) Proc Int Conf on Biological Effects of Low Dose Ionizing Radiation and Radioactive Pollutions on Environment, 17–21 September 2001, Syktyvkar, Russia, pp 254–255 (in Russian)
- Shevchenko VA, Abramov VI, Kalchenko VA, Fedotov IS, Rubanovich AV (1996) Genetic consequences of radioactive pollution of the environment caused by the Chernobyl accident for plants populations. Radiacionnaja biologija, Radioekologija 36;4:531–545 (in Russian)
- 39. Shevchenko VA, Kalchenko VA, Abramov VI, Rubanovich AV, Shevchenko VV, Grinikh LI (1999) Genetic effects in plants growing in the zone of the Kyshtym and Chernobyl accidents. Radiacionnaja biologija, Radioekologija 39;1:162–176 (in Russian)
- Sidorov VA (1990) Plant biotechnology. Cells selection. Naukova Dumka, Kiev (in Russian)
- Tautvydas KJ (1971) Mass isolation of pea nuclei. Plant Physiol 47:499–503
- 42. Underbrink AG, Sparrow RC, Sparrow AH, Rossi HH (1970) Relative biological effectiveness of X-rays and 0.43-MeV monoenergetic neutrons on somatic mutations and loss of reproductive integrity in *Tradescantia* stamen hairs. Radiat Res 44;1:187–203
- Urban PL, Bystrzejewska-Piotrowska G (2003) Cesium accumulation in plants and its ecophysiological effects. In: Proc of the 6th Int Symp & Exhibition on Environmental Contamination in Central and Eastern Europe and the Commonwealth of Independent States, 1–4 September 2003, Prague, Czech Republic. Manuscript 189, pp 132–136
- Wang W (1992) Use of plants for the assessment of environmental contaminants. Rev Environ Contam Toxicol 126:87–127
- 45. White PJ, Broadley MR (2000) Mechanisms of caesium uptake by plants. New Phytol 147:241–256
- 46. Zhu J, Gong Z, Zhang C et al. (2002) OSM 1/S4P61: a syntaxin protein in *Arabidopsis* controls abscisic acidmediated and non-abscisic acid-mediated responses to abiotic stress. Plant Cell 14:3009–3028
- Zhu YG, Smolders E (2000) Plant uptake of radiocaesium: a review of mechanisms, regulation and application. J Exp Bot 51:1635–1645